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(71) Applicant (*for all designated States except US*): **NATURAL AS** [NO/NO]; Kjoerdokollen 30, N-1337 Sandvika (NO).

(72) Inventors; and
(75) Inventors/Applicants (*for US only*): **REMMEREIT, Jan** [NO/NO]; Halkjelsgata 7, N-6100 Volda (NO). **KLAVENESS, Jo** [NO/NO]; Midtassen 5, N-1166 Oslo (NO).

(74) Agent: **OSLO PATENTKONTOR AS**; P.O. Box 7007 M, N-0306 Oslo (NO).

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(54) **Title:** COMPOSITIONS CONTAINING AMINOPOLYSACCHARIDES AND NEGATIVELY CHARGED POLYSACCHARIDES

(57) **Abstract:** The present invention relates to a readily dispersable composition that includes an aminopolysaccharide and a negatively charged polysaccharide. An aminopolysaccharide and a negatively charged polysaccharide such as xanthan gum, guar gum, alginic acid, carrageenan and pectin are blended together to form particles of chitosan coated with the polysaccharide. The composition may optionally contain food flavorings, dyes, vitamins, minerals, and/or phytonutrients.

COMPOSITIONS CONTAINING AMINOPOLYSACCHARIDES AND NEGATIVELY CHARGED POLYSACCHARIDES

FIELD OF THE INVENTION

The present invention relates to compositions including a positively charged polysaccharide (*e.g.*, aminopolysaccharide) and a negatively charged polysaccharide that are readily dispersable in aqueous solution. These compositions are suitable for use as dietary supplements.

BACKGROUND OF THE INVENTION

The beneficial effects of dietary fiber have long been recognized. An increase in dietary fiber has been recommended for a variety of indications, including reduction of serum cholesterol, reduction of the risk of coronary heart disease, reduction in blood pressure, enhanced weight control, better glycemic control, and reduced risk of cancers such as colon cancer. (*See, e.g.*, Anderson and Hanna, J. Nutr. 129(7 Suppl.):1457S-66S [1999]; Anderson *et al.*, Am. J. Clin. Nutr. 59(5 Suppl.):1242S-1247S [1994]; Jenkins *et al.*, J. Am. Coll. Nutr. 17(6):609-16 [1998]; Behall, Ann. N.Y. Acad. Sci. 819:142-54 [1997]). However, increasing the amount of fiber available in Western diets has been difficult.

One solution has been to provide dietary fiber as a supplement in bulk form. However, such supplements are often difficult to disperse in liquids and are unpalatable. For example, Stemmle *et al.* (European Pat. No. 080673, incorporated herein by reference) describes many of the problems associated with providing dietary fibers such as guar gum in bulk. When such fibers are provided in various forms (*e.g.*, tablets, capsules, powders), significant problems occur with respect to solubility, hydration, dispersion, and gelation. For example, tablets of guar gum are known to form a gel at their surface upon hydration, thereby effectively preventing dispersion of the fiber. Furthermore, powders made from dietary fiber are known to be difficult to disperse, often forming clumps of powder surrounded by a gel coating or a gelled, viscous mixture that is impossible or unpleasant to drink.

Several attempts have been made to solve these problems. For example, Applegren (U.S. Pat. No. 4,754,027, incorporated herein by reference) discloses using fatty acids, film-forming polymers, and ethyl cellulose to coat guar gum. However, the

resulting particle size causes a gritty texture and unpalatable mouthfeel, and the particles settle out when mixed in a solution. Likewise, Heath (G.B. Pat. No. 2,030,583, incorporated herein by reference) discloses a method for the granulation of guar gum by spraying with atomized water. However, the resulting particles have a gritty, unpleasant texture.

The dietary use of chitosan for the reduction of serum cholesterol and blood lipids has also been described. (*See*, *e.g.*, U.S. Pat. Nos. 4,223,023; 5,654,001; and 5,453,282; and European Pat. No. 775,450 A2, each of which is incorporated herein by reference). However, the recommended doses are between 3 and 12 grams before each meal. When chitosan is provided in tablet form, the recommended dose is equivalent to up to 12 tablets, which causes great inconvenience to the user. Furthermore, because chitosan only dissolves at acidic pH values, it does not disperse or dissolve in most liquids. Therefore, chitosan mixed in non-acidic liquids settles out rapidly and imparts a gritty, unpleasant texture and poor mouthfeel.

Accordingly, what is needed in the art are compositions containing sources of dietary fiber (*e.g.*, chitosan) and other carbohydrates that are dispersable in liquids, palatable, and have an acceptable mouthfeel. Preferably, the compositions are dispersable in the liquid at concentrations that provide an acceptable daily intake of the fiber or other carbohydrate.

SUMMARY OF THE INVENTION

The present invention relates to a readily dispersable composition that includes at least one positively charged polysaccharide (*e.g.*, and aminopolysaccharide) and at least one negatively charged polysaccharide. Accordingly, in some embodiments, the present invention provides a composition comprising a positively charged aminopolysaccharide and a negatively charged polysaccharide, wherein the composition is formed into a particle. In some embodiments of the present invention, the particle is dispersable in water. In other embodiments of the present invention, the particle binds substances having a log P of greater than about 1.5. In still other embodiments of the present invention, the aminopolysaccharide is selected from chitosan, chitosan derivatives, amino substituted cellulose, or combinations thereof. In further embodiments of the invention, the negatively charged polysaccharide is selected from xanthan, guar gum, carrageenan, pectin, alginic acid, and combinations thereof. In some particularly preferred

embodiments, the polysaccharide is xanthan. In some embodiments, the particle comprises greater than about 50% chitosan, while in other embodiments, the particle comprises greater than about 60% chitosan, 70% chitosan, 80% chitosan, 90% chitosan, or 99% chitosan. In some embodiments, the chitosan is a physiologically acceptable salt. In other embodiments, the particle further comprises a food additive selected from flavorings, colors, vitamins, minerals, fragrances, and phytonutrients. In some particularly preferred embodiments, the composition remains dispersed in a liquid after stirring. In some embodiments, the dispersion is a colloid. In still further embodiments, the particles further comprise an additional anionic compound (*e.g.*, DNA, RNA, or an anionic drug).

In other embodiments, the present invention provides a method comprising: a) providing an aminopolysaccharide, a negatively charged polysaccharide, and a liquid; b) combining the aminopolysaccharide, negatively charged polysaccharide, and liquid to form a mixture; and c) blending the mixture under conditions such that particles containing the aminopolysaccharide and negatively charged polysaccharide are formed. In some embodiments of the present invention, the particles bind compounds having a log P of greater than about 1.5. In some particularly preferred embodiments, the negatively charged polysaccharide is selected from xanthan, guar gum, carrageenan, pectin, and alginic acid. In some embodiments, the particle comprises greater than about 50% chitosan, while in other embodiments, the particle comprises greater than about 60% chitosan, 70% chitosan, 80% chitosan, 90% chitosan, or 99% chitosan. In some embodiments, the chitosan is a salt. In other embodiments, the particle further comprises a food additive selected from flavorings, colors, vitamins, minerals, fragrances, and phytonutrients. In some particularly preferred embodiments, the composition remains dispersed in a liquid after stirring.

DEFINITIONS

To facilitate understanding of the invention, a number of terms are defined below.

The term "chitosan" refers to co-polymers such as β-1$\rightarrow$4-linked 2-acetamido-2-deoxy-D-glucopyranose and β-1$\rightarrow$4-linked 2-amido-2-deoxy-D-glucopyranose formed by deacetylating chitin. It will be recognized that the level of deacetylation of chitin may vary so that the term "chitosan" encompasses any deacetylated chitin (*e.g.*, deacetylated

by at least forty percent to eighty percent, and in some instances to as much as ninety percent or ninety-five percent).

As used herein, the term "chitosan salt" refers to those salts formed by the reaction of chitosan and an acid, such as an organic acid or an inorganic acid (*e.g.*, as described in U.S. Pat. No. 5,599,916, incorporated herein by reference). Examples of chitosan salts formed with an inorganic acid include, but are not limited to, chitosan hydrochloride, chitosan hydrobromide, chitosan phosphate, and mixtures thereof. Examples of chitosan salts formed with an organic acid include, but are not limited to, salts of acetate adipate, benzene sulphonate, bromide, camsylate, chloride, citrate, glucuronate, hippurate, iodide, lactate, maleate, mesylate, napsulate, nitrate, oleate and other fatty acid anions including omega 3 and conjugated linoleic acid, as well as phosphate, succinate, sulphate, tartrate, and tosylate, and mixtures thereof. Chitosan salts produced using a mixture of acids including, for example, both inorganic and organic acids are also encompassed by this definition. Salts formed with nucleic acids such as DNA and anionic drugs such as COX-2 inhibitors, anionic steroid derivatives (*e.g.*, hydrocortisone hemisuccinate), and other drug anions for treatment of infections, cardiovascular diseases, diabetes, cancer, or asthma are also encompassed by the definition.

As used herein, the term "chitosan derivative" refers to derivatives formed by the reaction of chitosan with another chemical compound, including, but not limited to compounds such as aromatic aldehydes (*e.g.*, salicylaldehyde, 3-formyl-2-hydroxybenzoic acid, pyridine aldehydes, etc.), aldoses and ketoses (*e.g.*, glucose, galactose, arabinose, xylose, N-acetylglucosamine, lactose and maltose) so that a chemical group not normally present in chitosan is covalently attached to the chitosan molecule. Chemical modifications creating chitosan derivatives can in general involve any chemical modification of one or more alcohol groups or one or more amino groups. These groups can be removed and replaced with substitute groups and/or optionally reacted with chemical groups in other compounds or directly reacted with other organic molecules. Typical examples are O-alkylation forming an ether linkage, O-acylation forming an ester, O-alkylation forming acetals, N-alkylation forming amines, and N-acylation forming amides. The derivitives of chitosan can be more hydrophilic or more lipophilic than the parent molecule. Furthermore, the derivatives can be anionic, cationic, or neutral.

As used herein, the term "chitosan containing material" refers to materials comprising chitosan, chitosan salts, chitosan derivatives, and mixtures thereof.

As used herein, the term "polysaccharide" refers to a polymer comprising monosaccharides arranged in monomeric units (*e.g.*, homoglycans, diheteroglycans, triheteroglucans, tetraheteroglycans, pentaheteroglucans) and includes both naturally occurring polysaccharides, derivatives of polysaccharides, and extracts from seaweed, seeds, tubers and roots. The term "polysaccharide" applies to all classes of polysaccharides, including linear polysaccharides (*e.g.*, algins, carrageenans, galactomannans [*e.g.*, guar gum and locust bean gum, and pectins], branched polysaccharides [*e.g.*, arabinans, arabinoglactans, xanthan, and xylans], and branch-on-branch polysaccharides [*e.g.*, amylopectins, arabinoxylans]). Examples of polysaccharides include, but are not limited to, xanthan, agar, alginic acid, carrageenans, pectins, corn starch, rice starch, wheat starch, guar gum, locust bean gum, psyllium seed gum, potato starch, tapioca starch, gum arabic, gum karaya, and cellulose.

As used herein, the term “aminopolysaccharide,” refers to positively charged polysaccharides including, but not limited to, chitosan and chitosan derivatives (*e.g.*, N-alkylated chitosan derivatives), amino substituted cellulose, and amino-substituted starch.

As used herein, the term "carrageenan" refers to the group of sulfated galactans (*e.g.*, kappa-carrageenan and lambda-carrageenan) isolated from red seaweeds.

As used herein, the term "soluble polysaccharide" refers to polysaccharides that are dissolvable in water or another liquid. Examples of soluble polysaccharides include, but not limited to, xanthan, agar, alginic acid, carrageenans, pectins, corn starch, rice starch, wheat starch, guar gum, locust bean gum, psyllium seed gum, potato starch, tapioca starch, gum arabic, and gum karaya.

As used herein, the terms "anionic polysaccharide" or “negatively charged polysaccharide” refer to acidic polysaccharides or polysaccharides having a net negative charge (*e.g.*, polysaccharides containing the following groups: uronic ester, sulfate half-ester, pyruval cyclic acetal, succinate half-ester) including, but not limited to, xanthan, xylans, pectins, carrageenans, algins, arabinoxylans, psyllium seed gum, gum arabic, and fucellarans.

As used herein, the term "neutral polysaccharide" refers to polysaccharides having a neutral charge and includes, but is not limited to, amylopectins, amyloses, arabinans, guar gum and other galactomannans, and arabinogalactans.

As used herein, the term "dietary fiber" refers to nondigestable polysaccharides, including, but not limited to, xanthan, agar, alginic acid, carrageenans, pectins, guar gum, locust bean gum, psyllium seed gum, gum arabic, gum karaya, and cellulose.

As used herein, the term "soluble dietary fiber" refers to nondigestable polysaccharides that are dissolvable in water, including, bit not limited to, xanthan, agar, alginic acid, carrageenans, pectins, guar gum, locust bean gum, psyllium seed gum, gum arabic, and gum karaya.

As used herein, the term “partition coefficient” or “P” is used herein as a measure of lipophilicity. The partition coefficient is defined by the following formula:

$$P = \frac{[\text{compound}]_{oct}}{[\text{compound}]_{aq}(1 - a)}$$

wherein a is the degree of dissociation of the compound in water calculated from ionization constants, $[\text{compound}]_{oct}$ is concentration in octanol, and $[\text{compound}]_{aq}$ is concentration in water. The partition coefficient may be derived experimentally by placing a compound in shaking device (*e.g.*, a separatory funnel) with varying volumes of octanol and water, determining the concentration of the compound in each layer after mixing, and utilizing the equation to calculate P. In general, the more lipophilic the compound, the higher the partition coefficient.

As used herein, the term "log P" is also used to refer to the lipophilicity of a compound and is the log of the partition coefficient. It is known that the rate of movement of a variety of organic compounds through cellular material is approximately proportional to the logarithm of partitional coefficients between an organic solvent and water. The relative potency of a drug, expressed as log I/C, where C is the concentration of the drug that produces some standard biologic effect, is related to lipophilicity by the following equation:

$$\log 1/C = -k(\log P)^2 + k'(\log P) + k''$$

On the basis of this equation, it is apparent that if a compound is more soluble in water than in octanol, P is less than 1, and, therefore, log P is negative. Conversely, a molecule more soluble in 1-octanol has a P value greater than 1, and log P is positive. The larger the value of P, the more there will be an interaction of the drug with the lipid phase (*e.g.*, membranes). As P approaches infinity, the drug interaction will become so great that the drug will not be able to cross the aqueous phase, and it will localize to the first lipophilic phase with which it comes into contact. As P approaches zero, the drug

will be so water soluble that it will not be capable of crossing the lipid phase and will localize in the aqueous phase. Somewhere between P = 0 and P = infinity there will be a value of P such that drugs having this value will be least hindered in their journey through macromolecules to the site of action. This value is called log P_o, the optimum partition coefficient for biological activity.

As used herein, the term "particle" refers to a small piece of matter.

As used herein, the term "dispersable" refers to ability of compound to scatter or disaggregate in a liquid.

As used herein, the term "food additive" refers to any material approved by the United States Department of Agriculture for inclusion in foodstuffs and substances that are generally recognized as safe (GRAS) and includes all substances, the intended use of which results or may reasonably be expected to result, directly or indirectly, either in their becoming a component of food or otherwise affecting the characteristics of food.

As used herein, the term "food flavoring" refers to food additive or other substance derived from an animal, mineral, plant, or synthetic source capable of imparting a flavor to a substance or changing the flavor of a substance.

As used herein, the term "food coloring" refers to any dye, pigment, or other substance made by a process of synthesis or similar artifice, or extracted, isolated, or otherwise derived, with or without intermediate or final change of identity, from a vegetable, animal, mineral, or other source and that, when added or applied to a food, drug, or cosmetic or to the human body or any part thereof, is capable (alone or through reaction with another substance) of imparting a color thereto or changing the color thereof.

As used herein, the term "vitamin" refers to an organic molecule that is essential for the health of an animal, and includes, but is not limited to, vitamin A, thiamin (B1), riboflavin (B2), Pyridoxine (B6), cyanocobalamin (B12), biotin, ascorbic acid (vitamin C), retinoic acid (vitamin D), vitamin E, folic acid and other folates, vitamin K, niacin, and pantothenic acid and their derivatives.

As used herein, the term "mineral" refers to elements (*e.g.*, sodium, potassium, magnesium, calcium, phosphorus, and chlorine) and trace elements (*e.g.*, iron, zinc, manganese, fluorine, copper, molybdenum, chromium, selenium, and iodine) required for normal body function.

As used herein, the term "phytonutrient" refers to organic compounds isolated from plants that have a biological effect, and includes, but is not limited to, compounds of the following classes: isoflavonoids, oligomeric proanthcyanidins, indol-3-carbinol, sulforaphone, fibrous ligands, plant phytosterols, ferulic acid, anthocyanocides, triterpenes, omega 3/6 fatty acids, polyacetylene, quinones, terpenes, cathechins, gallates, and quercitin.

DESCRIPTION OF THE INVENTION

The present invention relates to compositions including an aminopolysaccharide and a negatively charged polysaccharide that are readily dispersable in aqueous solution and find use as dietary supplements and in foods. The particles of the present invention have desirable organoleptic properties (*e.g.*, good mouth feel) and are useful for binding both dietary and circulating lipids in the body. A variety of additional substances may be incorporated into or used in conjunction with the particles, including, but not limited to vitamins, minerals, antioxidants, phytonutrients, food colorings, flavorings, preservatives, powdered food products, freeze dried food products, and whole food products.

A. Aminopolysaccharide and Negatively Charged Polysaccharide Particles

The present invention provides particles comprising an aminopolysaccharide (*e.g.*, chitosan, one of its derivatives, or amino-substituted cellulose, or mixtures thereof) and a negatively charged polysaccharide. In some embodiments of the present invention, the chitosan is a co-polymer of β-1→4-linked 2-acetamido-2-deoxy-D-glucopyranose and β-1→4-linked 2-amido-2-deoxy-D-glucopyranose formed by the alkaline deacetylation of chitin with concentrated sodium hydroxide at elevated temperatures (*See*, *e.g.*, Varum *et al.*, Carbohydrate Res. 211:17-23 [1991]; Varum *et al.*, Carbohydrate Res. 217:19-27 [1991]). As is known in the art, the degree of deacetylation may vary. In some embodiments of the present invention, the chitosan is deacetylated to between about forty percent and ninety percent. In other embodiments of the present invention, the particles comprise one or more derivatives of chitosan. In some embodiments, the derivative is a chitosan salt (*e.g.*, chitosan hydrochloride, chitosan hydrobromide, chitosan malate, chitosan phosphate, chitosan formate, chitosan acetate, chitosan propionate, chitosan

chloroacetate, chitosan hydroxyacetate, chitosan butyrate, chitosan isobutyrate, chitosan acrylate, and mixtures thereof). In other embodiments, the derivative is a covalently modified chitosan derivative (*e.g.*, carboxymethylchitosan, N-salicylidenechitosan, succinylchitosan, N-carboxyacylchitosan, etc.).

The particles of the present invention also comprise one or more of a variety of polysaccharides. In some particularly preferred embodiments, the polysaccharide is an anionic or negatively charged polysaccharide (*e.g.*, xanthan, xylans, pectins, carrageenans, algins, arabinoxylans, psyllium seed gum, gum arabic, and fucellarans, or mixtures thereof). In other embodiments, the polysaccharide is a neutral polysaccharide (*e.g.*, amylopectins, amyloses, arabinans, guar gum and other galactomannans, and arabinogalactans, or mixtures thereof).

The chitosan and polysaccharide particles of the present invention have a variety of formulations. In each case, the formulation provides a particle that is dispersable in aqueous solutions. Furthermore, the resulting dispersion is pleasant to drink, with an acceptable mouthfeel and taste. In some embodiments, the concentration of chitosan is about 50% to 99%, while the concentration of polysaccharide is about 1% to 50%. In some particularly preferred embodiments, the concentration of chitosan is from about 60% to 80%, while the concentration of polysaccharide is from about 20% to 40%. In some embodiments, the particles binds compounds having a log P of greater than 1.5. In still further embodiments, the particles comprise one or more additional compounds as described below. In other embodiments of the present invention, a plurality of particles are provided as a powder, or alternatively, in capsules, tablets, or liposomes.

B. Additional Compounds for Inclusion in Particles

It is contemplated that variety of compounds and substances may be incorporated into the aminopolysaccharide and negatively charged polysaccharide particles of the present invention. These compounds and substances may add to the palatability or sensory perception of the particles (*e.g.*, flavorings and colorings) or improve the nutritional value of the particles (*e.g.*, minerals, vitamins, phytonutrients, antioxidants, etc.). In some embodiments, the included compounds have a log P of greater than 1.5. In other embodiments, the particles comprise physiological acceptable salts (*e.g.*, $NaCl_2$, $CaCl_2$, $MgSO_4$) and non-ionic physiologically acceptable low molecular weight compounds (*e.g.*, glucose).

In further embodiments, the particles comprise at least one food flavoring such as acetaldehyde (ethanal), acetoin (acetyl methylcarbinol), anethole (parapropenyl anisole), benzaldehyde (benzoic aldehyde), N-butyric acid (butanoic acid), d- or l-carvone (carvol), cinnamaldehyde (cinnamic aldehyde), citral (2,6-dimethyloctadien-2,6-al-8, gera-nial, neral), decanal (N-decylaldehyde, capraldehyde, capric aldehyde, caprinaldehyde, aldehyde C-10), ethyl acetate, ethyl butyrate, 3-methyl-3-phenyl glycidic acid ethyl ester (ethyl-methyl-phenyl-glycidate, strawberry aldehyde, C-16 aldehyde), ethyl vanillin, geraniol (3,7-dimethyl-2,6 and 3,6-octadien-1-ol), geranyl acetate (geraniol acetate), limonene (d-, l-, and dl-), linalool (linalol, 3,7-dimethyl-1,6-octadien-3-ol), linalyl acetate (bergamol), methyl anthranilate (methyl-2-aminobenzoate), piperonal (3,4-methylenedioxy-benzaldehyde, heliotropin), vanillin, alfalfa (*Medicago sativa L.*), allspice (*Pimenta officinalis*), ambrette seed (*Hibiscus abelmoschus*), angelic (*Angelica archangelica*), Angostura (*Galipea officinalis*), anise (*Pimpinella anisum*), star anise (*Illicium verum*), balm (*Melissa officinalis*), basil (*Ocimum basilicum*), bay (*Laurus nobilis*), calendula (*Calendula officinalis*), (*Anthemis nobilis*), capsicum (*Capsicum frutescens*), caraway (*Carum carvi*), cardamom (*Elettaria cardamomum*), cassia, (*Cinnamomum cassia*), cayenne pepper (*Capsicum frutescens*), Celery seed (*Apium graveolens*), chervil (*Anthriscus cerefolium*), chives (*Allium schoenoprasum*), coriander (*Coriandrum sativum*), cumin (*Cuminum cyminum*), elder flowers (*Sambucus canadensis*), fennel (*Foeniculum vulgare*), fenugreek (*Trigonella foenum-graecum*), ginger (*Zingiber officinale*), horehound (*Marrubium vulgare*), horseradish (*Armoracia lapathifolia*), hyssop (*Hyssopus officinalis*), lavender (*Lavandula officinalis*), mace (*Myristica fragrans*), marjoram (*Majorana hortensis*), mustard (*Brassica nigra, Brassica juncea, Brassica hirta*),
nutmeg (*Myristica fragrans*), paprika (*Capsicum annuum*), black pepper (*Piper nigrum*), peppermint (*Mentha piperita*), poppy seed (*Papayer somniferum*), rosemary (*Rosmarinus officinalis*), saffron (*Crocus sativus*), sage (*Salvia officinalis*), savory (*Satureia hortensis, Satureia montana*), sesame (*Sesamum indicum*), spearmint (*Mentha spicata*), tarragon (*Artemisia dracunculus*), thyme (*Thymus vulgaris, Thymus serpyllum*), turmeric (*Curcuma longa*), vanilla (*Vanilla planifolia*), zedoary (*Curcuma zedoaria*), sucrose, glucose, saccharin, sorbitol, mannitol, aspartame. Other suitable flavoring are disclosed in such references as Remington's Pharmaceutical Sciences, 18th Edition, Mack Publishing, p.

1288-1300 [1990], and Furia and Pellanca, Fenaroli's Handbook of Flavor Ingredients, The Chemical Rubber Company, Cleveland, Ohio, [1971], known to those skilled in the art.

In other embodiments, the particles comprise at least one synthetic or natural food coloring (*e.g.*, annatto extract, astaxanthin, beet powder, ultramarine blue, canthaxanthin, caramel, carotenal, beta carotene, carmine, toasted cottonseed flour, ferrous gluconate, ferrous lactate, grape color extract, grape skin extract, iron oxide, fruit juice, vegetable juice, dried algae meal, tagetes meal, carrot oil, corn endosperm oil, paprika, paprika oleoresin, riboflavin, saffron, tumeric, tumeric and oleoresin).

In still further embodiments, the particles comprise at least one phytonutrient (*e.g.*, soy isoflavonoids, oligomeric proanthcyanidins, indol-3-carbinol, sulforaphone, fibrous ligands, plant phytosterols, ferulic acid, anthocyanocides, triterpenes, omega 3/6 fatty acids, polyacetylene, quinones, terpenes, cathechins, gallates, and quercitin). Sources of plant phytonutrients include, but are not limited to, soy lecithin, soy isoflavones, brown rice germ, royal jelly, bee propolis, acerola berry juice powder, Japanese green tea, grape seed extract, grape skin extract, carrot juice, bilberry, flaxseed meal, bee pollen, ginkgo biloba, red clover, burdock root, dandelion, parsley, rose hips, milk thistle, ginger, Siberian ginseng, rosemary, curcumin, garlic, lycopene, grapefruit seed extract, spinach, and broccoli.

In still other embodiments, the particles comprise at least one vitamin (*e.g.*, vitamin A, thiamin (B1), riboflavin (B2), pyridoxine (B6), cyanocobalamin (B12), biotin, ascorbic acid (vitamin C), retinoic acid (vitamin D), vitamin E, folic acid and other folates, vitamin K, niacin, and pantothenic acid). In some embodiments, the particles comprise at least one mineral (*e.g.*, sodium, potassium, magnesium, calcium, phosphorus, chlorine, iron, zinc, manganese, flourine, copper, molybdenum, chromium, selenium, and iodine). In some particularly preferred embodiments, a dosage of a plurality of particles includes vitamins or minerals in the range of the recommended daily allowance (RDA) as specified by the United States Department of Agriculture. In still other embodiments, the particles comprise an amino acid supplement formula in which at least one amino acid is included (*e.g.*, l-carnitine or tryptophan).

C. Manufacture of Particles

The aminopolysaccharide and negatively charged polysaccharide compositions are made by combining varying amounts of an aminopolysaccharide and a negatively charged polysaccharide. It is contemplated that those skilled in the art understand that various specific formulations of aminopolysaccharide and negatively charged polysaccharide particles described below can be altered or modified without changing the properties (*e.g.*, partition coefficient, dispersability, fat binding capacity, mouthfeel when dispersed in aqueous solution) of the particles.

In some embodiments of the present invention, an aminopolysaccharide (*e.g.*, chitosan or a derivative of chitosan), water (or an aqueous solution), and a negatively charged polysaccharide (*e.g.*, xanthan) are mixed together to form a slurry. Generally, an amount of aminopolysaccharide is chosen so that the final composition (*i.e.*, particle) will comprise about 0.1% to 99.9% aminopolysaccharide on a weight/weight (w/w) basis, preferably about 50% to 90% aminopolysaccharide (w/w), and most preferably about 60% to 80% (w/w) aminopolysaccharide, and be able to bind compounds having a log P of greater than about 1.5. The amount of the negatively charged polysaccharide is varied in relation to the aminopolysaccharide such that final composition (*i.e.*, particle) comprises about 0.1% to 99.9% negatively charged polysaccharide on weight/weight (w/w) basis, preferably about 10% to 50% negatively charged polysaccharide (w/w), and most preferably about 20% to 40% (w/w) negatively charged polysaccharide.

The amount of water added to the slurry may also vary. Generally, enough water is added to wet the aminopolysaccharide. In some embodiments, about 10% to 50% water is added on weight/volume (w/v) basis. In some particularly preferred embodiments, about 30% to 40% (w/v) water is added. In still other embodiments, the dry aminopolysaccharide and negatively charged polysaccharide are mixed, and then water is added. In still further embodiments where an aqueous solution is utilized, the solution can contain food flavorings, colorings, phytonutrients, vitamins, and minerals as described above. In some embodiments, one or more negatively charged polysaccharides (*e.g.*, xanthan or guar gum) are added to the aminopolysaccharide slurry and the mixture is blended vigorously until particles form. In still further embodiments, water is sprayed or added to a dry mixture of chitosan and polysaccharide while blending. The amount of polysaccharide added may vary, but it should be sufficient to cause particle formation.

Any suitable blending apparatus may be used to blend or mix the aminopolysaccharide and the negatively charged polysaccharide. For example, in some embodiments, the mixture is blended in a high speed Waring blender. In some particularly preferred embodiments, the mixture is mixed in an Ide-Con (Porsgrunn, Norway) mixing apparatus. In still further embodiments, the mixture is blended in the blending apparatus until particles of the desired size, shape, and properties form. In other embodiments, blending is continued until the particles are dried. In some particularly preferred embodiments, the aminopolysaccharide and water are blended for about an 0.5 to 1.0 hours, the negatively charged polysaccharide is added, and the blending continued until the mixture dries. In other embodiments, the mixture is blended until particles of about 1 micron to 5 millimeters are formed. In some particularly preferred embodiments, the particles are dried while mixing at about 50°C to 80°C, preferably about 60°C. Additional compounds (*e.g.*, food flavorings, colorings, phytonutrients, vitamins, and minerals as described above) may be added to the particles before or, preferably, after drying.

In still further embodiments, other agents are utilized to form coated chitosan particles. In some embodiments, chitosan is coated with gelatin or xanthan, while in other embodiments, chitosan is coated with glycerol. In each instance, an amount of the coating agent sufficient to coat the chitosan is added to the chitosan, and the mixture blended vigorously as described above until the desired particles form. In some embodiments, a glycerol solution is sprayed onto chitosan particles. In some preferred embodiments, the solution is about 20% to 50% glycerol on a volume/volume (v/v) basis. In other preferred embodiments, an amount of glycerol is chosen so that the final composition (*i.e.*, particle) will comprise about 10% to 50% (w/w) glycerol, preferably about 15% to 30% (w/w) glycerol. In other embodiments, a gelatin solution is added to a chitosan and the mixture mixed vigorously until particles form. In some embodiments, an amount of gelatin is chosen so that the final composition (*i.e.*, particle) will comprise about 5% to 50% (w/w) gelatin, preferably about 15% to 30% (w/w) gelatin.

It is not intended that the present invention be limited to particular mechanism of action. Indeed, an understanding of the mechanism is not necessary to make and use the present invention. However, it is believed that the process produces a particle comprising a chitosan core with a polysaccharide coating. When a negatively charged polysaccharide is utilized, the polysaccharide is attracted to the positively charged

chitosan. When the particles are added to water or an aqueous solution, the particles disperse readily due to hydration of the polysaccharide coating. Indeed, it is contemplated that in some cases the resulting composition is colloidal in nature. It is also contemplated that hydration of the polysaccharide coating eliminates or reduces the gritty mouthfeel of uncoated chitosan.

In other embodiments, the present invention provides chitosan and polysaccharide particles comprising a polysaccharide core coated with chitosan and/or a chitosan derivative. In some embodiments, a polysaccharide is coated with chitosan by vigorously blending the polysaccharide with a chitosan solution. In some embodiments, the chitosan solution comprises an aqueous solution of chitosan and/or a chitosan derivative. It is well known in the art that chitosan is insoluble in neutral and basic aqueous solutions. Accordingly, it is contemplated that the pH of the chitosan solution be acidic. In some embodiments, the solution is acidified with lactic acid. In some particularly preferred embodiments, the chitosan solution comprises about 1% to 20% (w/v) chitosan, preferably about 2% chitosan. In other embodiments, the chitosan solution comprises about 1% to 10% (w/v) lactic acid, preferably about 4% (w/v) lactic acid. It is further contemplated that a sufficient amount of the chitosan solution is combined with the polysaccharide so that particles are formed during vigorous mixing. In some embodiments, about 0.1 to 5 ml of chitosan solution is added per gram of polysaccharide. In some particularly preferred embodiments, about 0.5 to 1.0 ml of chitosan solution is added per gram of polysaccharide. In still further embodiments, additional compounds (*e.g.*, food flavorings, colorings, phytonutrients, vitamins, and minerals as described above) are incorporated into the solution or added before or after mixing. In other embodiments, the particles are dried after formation.

D. Dietary Use of Chitosan and Polysaccharide Particles

The present invention contemplates the dietary use of the aminopolysaccharide and negatively charged polysaccharide particles. Because the particles are readily dispersable in aqueous solution and have an improved mouthfeel as compared to chitosan alone, dosages of from 0.1 to 100 grams, preferably about 1 to 20 grams, and most preferably about 3 to 12 grams, can be consumed. The present invention is not limited to any particular timing of consumption. A dose of the particles may be consumed

weekly, daily, or several times throughout the day (*e.g.*, before or after one or several daily meals).

It is not intended that the present invention be limited to particular mechanism of action. Indeed, an understanding of the mechanism is not necessary to make and use the present invention. However, it is contemplated that the particles of the present invention have certain biological effects following ingestion. In particular, the chitosan and polysaccharide particles bind fatty acids and other dietary and circulating lipids (*e.g.*, triglycerides, bile acids, cholesterol, and other sterols). Furthermore, ingestion of the particles of the present invention leads to increase in the amount of lipid excreted in the faeces. Accordingly, particles of the present invention find use as a pharmaceutical for the treatment gall bladder disease, in which the absorption and utilization of lipids is not desirable; in the treatment of obesity; and in gastro-intestinal disorders or coronary disease where bile acids, cholesterol, and other sterol lowering is desired. Thus, in some embodiments of the present invention, it is contemplated that a biologically or therapeutically effective amount of the particles of the present invention is that amount that upon use for a period of about three months, causes a decrease in circulating lipids. In other embodiments, it is contemplated that a biologically or therapeutically effective amount is that amount that causes an increase in the lipid content of the stool. In still further embodiments, the particles find use in enhancing the excretion of exogenous materials, toxins, and drugs from the body. For example, it is contemplated that oral intake of the particles will enhance excretion of waste products in patients with reduced kidney function.

In some embodiments, it is also contemplated that particles of the present invention find use for inclusion in dietary supplement powders and drinks, including those formulated for use in very low calorie diets. Examples of such drinks and supplements are described in U.S. Pat. Nos. 5,470,839; 5,760,082; 4,282,265; 5,904,924; 5,948,460; 4,814,172; 4,834,990; each of which is incorporated herein by reference. It is contemplated that these formulations may be altered by substituting the fiber component with the particles of the present invention, or simply adding particles to the formulations.

The particles of the present invention also find use as carriers of pharmaceutical compositions (*e.g.*, anionic drugs). In some preferred embodiments, the pharmaceutical composition is incorporated into the particle before or during blending. Accordingly, the particles may be provided alone or in combination with any of the compounds described

above in tablets, pills, dragees, capsules, solutions, liquids, slurries, liposomes, suspensions and emulsions. The particles and compounds may be further provided in aqueous solution, oily solution, as a powder, or in any of the other forms discussed above. The tablet or capsule of the present invention may be coated with an enteric coating which dissolves at a pH of about 6.0 to 7.0. A suitable enteric coating which dissolves in the small intestine but not in the stomach is cellulose acetate phthalate. Further details on techniques for formulation for and administration and administration may be found in the latest edition of *Remington's Pharmaceutical Sciences* (Maack Publishing Co., Easton, PA).

EXPERIMENTAL

The following examples are provided in order to demonstrate and further illustrate certain preferred embodiments and aspects of the present invention and are not to be read as limiting the scope thereof.

Example 1

This Example describes the synthesis of polysaccharide coated chitosan particles. Thirty grams of deodorized shrimp chitosan (Natural Biopolymer, Inc./Vanson, Raymond, WA) is sprayed with a 12 ml water in which 0.07 grams vanilla (SFK Norge AS, Skytta, Norway) and 0.5 grams of aspartam have been dissolved. Next, 13.5 grams of xanthan gum (SFK Norge) is added to slurry while mixing in an Ide-Con mixer (Porsgrun, Norway) for approximately one hour to form particles. The particles are then dried with hot air (60°C) while mixing. After the particles are dried, 0.5 grams of blood orange powder (Ringe & Kuhlmann, Hamburg, Germany) is added. The composition (wt%) after drying is:

Deodorized shrimp chitosan:	64.0
Water:	5.0
Vanilla:	0.15
Aspartam:	1.0
Xanthan gum:	28.8
Blood orange powder:	1.0

It will be recognized the process described above may be altered by substituting guar gum, carrageenan, alginic acid, pectin and other polysaccharides for the xanthan gum. Likewise, chitosan salts and other derivatives of chitosan may replace the deodorized shrimp chitosan. Furthermore, other food additives such as sweeteners, flavorings, vitamins, minerals, phytonutrients may be added before, after, or during mixing and drying.

Example 2

This Example describes the synthesis of polysaccharide coated chitosan particles. Thirty grams of deodorized shrimp chitosan is sprayed with a 12 ml water in which 0.07 grams vanilla and 0.5 grams of aspartame have been dissolved. Next, 13.5 grams of xanthan gum is added to slurry while mixing in an Ide-Con mixer (Porsgrun, Norway) for approximately one hour to form particles. The particles are then dried with hot air (60°C) while mixing. After the particles are dried, 1.0 grams of blood orange powder is added.

Example 3

This Example describes the synthesis of polysaccharide coated chitosan particles. Thirty grams of deodorized shrimp chitosan, 10 grams of xanthan gum, and 0.4 grams of aspartam are mixed by shaking in an Erlenmeyer flask. The mixture is transferred to an Ide-Con mixer (Porsgrun, Norway), and 42 ml water is sprayed into the mixture and 0.2 grams blood orange powder is added while vigorously mixing for about one hour. The particles are then dried with hot air (50-60°C) while mixing.

It will be recognized the process described above may be altered by substituting guar gum, carrageenan, alginic acid, pectin and other polysaccharides for the xanthan gum. Likewise, chitosan salts and other derivatives of chitosan may replace the deodorized shrimp chitosan. Furthermore, other food additives such as sweeteners, flavorings, vitamins, minerals, phytonutrients may be added before, after, or during mixing and drying.

Example 4

This Example describes the preparation of polysaccharide coated chitosan particles. Thirty grams of deodorized shrimp chitosan, 10 grams of lactic acid powder, 9.5 grams xanthan gum, and 0.5 grams of aspartame, and 0.025 grams blood orange powder are mixed by shaking in an Erlenmeyer flask. The mixture is transferred to an Ide-Con mixer (Porsgrun, Norway), and 70 ml water is sprayed into the mixture vigorously mixing and drying with hot air (50-60°C) for about one hour. During mixing, another 0.23 grams of blood orange powder are added. The particles are then dried with hot air (50-60°C) while mixing.

It will be recognized the process described above may be altered by substituting guar gum, carrageenan, alginic acid, pectin and other polysaccharides for the xanthan gum. Likewise, chitosan salts and other derivatives of chitosan may replace the deodorized shrimp chitosan. Furthermore, other food additives such as sweeteners, flavorings, vitamins, minerals, phytonutrients may be added before, after, or during mixing and drying.

Example 5

This Example describes the coating of xanthan gum with chitosan. Thirty grams of xanthan gum in a mixing bowl is sprayed with 16 ml of a filtered water solution containing 2% by weight deodorized shrimp chitosan and 4% by weight lactic acid while mixing vigorously. The particles are then dried with hot air (50-60°C) while mixing.

Example 6

This Example describes the coating of xanthan gum with chitosan. Thirty grams of xanthan gum in a mixing bowl is sprayed with 20 ml of a filtered water solution containing 2% by weight deodorized shrimp chitosan, 4% by weight lactic acid, and 0.3 grams aspartame, while mixing vigorously. The particles are then dried with hot air (50-60°C) while mixing.

It will be recognized the process described above may be altered by substituting guar gum, carrageenan, alginic acid, pectin and other polysaccharides for the xanthan

gum. Likewise, chitosan salts and other derivatives of chitosan may replace the deodorized shrimp chitosan. Furthermore, other food additives such as sweeteners, flavorings, vitamins, minerals, phytonutrients may be added before, after, or during mixing and drying.

Example 7

This Example describes the coating of chitosan particles with glycerol. Glycerol (15.5 grams) and 15 ml water are mixed into a homogenous mixture and heated to 60°C. The glycerol/water mixture is then sprayed onto 65 grams of deodorized shrimp chitosan under vigorous mixing conditions and while continuously drying with hot air (50-60°C).

Example 8

This Example describes the coating of chitosan particles with gelatin. Forty grams of deodorized shrimp chitosan and 10 grams of lactic acid powder are mixed under vigorous conditions. A gelatin solution (7.5 grams gelatin dissolved into 100 ml water at 60°C) is then sprayed into the chitosan mixture while vigorously mixing and drying with hot air (50-60°C).

Example 9

Examples 9-12 are *in vitro* experiments stimulating fat binding properties *in vivo*. The particles are combined with fat under acidic pH (simulating gastric conditions) followed by treatment at neutral conditions (simulating intestinal conditions). It is assumed that compounds that are not extractable with chloroform are not available to or extractable by the body. This example compares the fat binding capacities of the chitosan/xanthan gum particles prepared essentially as described above in Examples 1-4 and chitosan alone. Soybean oil (5.0 g) and chitosan/xanthan gum particles (5.0 g) are dissolved in a stirred aqueous solution (0.1 N HCL, 500 ml) at 37°C. Following dissolution, the pH of the solution is adjusted to about 7 to 8 by the careful addition of sodium hydroxide pellets. The mixture is stirred for 2 hours at 37°C and subjected to high vacuum filtration to separate gelled solution from non-gelled solution. The

separated gel and solution are then extracted with chloroform (3 x 200 ml) and then dried with magnesium sulfate, filtered, evaporated, and weighed. The data indicate that the chitosan/xanthan particles bind the oil more tightly than chitosan alone.

Table 1

Fat Binding Capacity

Fat Binding Agent	Viscosity	Oil Recovered from Gel	Oil recovered from Solution
Chitosan/xanthan particles	High	2.70 g	0.05 g
Chitosan	High	3.98 g	0.10 g
None	Low	-	5.01 g

Example 10

This example compares the fat binding capacities of chitosan/xanthan gum particles prepared essentially as described in Examples 1-4 and chitosan alone. This experiment is performed as in Example 9, except that 50 g of soybean oil is mixed with 5.0 g fat binding agent in 1000 ml aqueous solution (0.1 N HCL), and the mixture is extracted three times in 300 ml chloroform. No filterable gel is formed. The data is presented in Table 2. As in Example 9, the xanthan/chitosan particles appear to have a higher affinity for soybean oil as compared to chitosan alone.

Table 2 Fat Binding Capacity	
Fat Binding Agent	**Fat Extracted with Chloroform**
Chitosan/xanthan particles	21.3 g
Chitosan	39.3 g
None	51.0 g

Example 11

This example compares the fat binding capacities of chitosan/xanthan gum particles prepared essentially as described in Examples 1-4 and chitosan alone. This experiment is performed as in Example 9, except that 5.0 g palmitic acid is mixed with 5.0 g fat binding agent. No filterable gel is formed by the chitosan/xanthan particles. The data is presented in Table 2. As in Example 9, the xanthan/chitosan particles appear to have a higher affinity for soybean oil as compared to chitosan alone.

Table 3 Fat Binding Capacity			
Fat Binding Agent	**Viscosity**	**Palmitic Acid Extracted from Gel**	**Palmitic Acid Extracted from Solution**
Chitosan/xanthan particles	Low	-	2.74 g
Chitosan	High	4.2 g	0.06 g

Example 12

This example compares the fat binding capacities of chitosan/xanthan gum particles prepared essentially as described in Examples 1-4 and chitosan alone. This experiment is performed as in Example 9, except that 10 g of palmitic acid is mixed with 1.0 g fat binder in 1000 ml aqueous solution (0.1 N HCL), and the mixture is extracted three times in 300 ml chloroform. No filterable gel is formed. The data is presented in Table 4. As in Example 9, the xanthan/chitosan particles appear to have a higher affinity for soybean oil as compared to chitosan alone.

Table 4
Fat Binding Capacity

Fat binder	Viscosity	Palmitic Acid Extracted with Chloroform
Chitosan/xanthan particles	Low	7.3 g
Chitosan	Low	13.2 g

What should be clear from above is that the present invention provides compositions comprising particles of an aminopolysaccharide and a negatively charged polysaccharide, and optionally one or more food additives. The particles are readily dispersable in liquids, do not settle rapidly, and have a pleasant mouthfeel.

All publications and patents mentioned in the above specification are herein incorporated by reference. Various modifications and variations of the described method and system of the invention will be apparent to those skilled in the art without departing from the scope and spirit of the invention. Although the invention has been described in connection with specific preferred embodiments, it should be understood that the invention as claimed should not be unduly limited to such specific embodiments. Indeed, various modifications of the described modes for carrying out the invention which are obvious to those skilled in food science, medicine, biochemistry, or related fields are intended to be within the scope of the following claims.

CLAIMS

What is claimed is:

1. A composition comprising a positively charged aminopolysaccharide and a negatively charged polysaccharide, wherein said composition is formed into a particle.

2. The composition of Claim 1, wherein said particle is dispersable in water.

3. The composition of Claim 1, wherein said particle binds compounds having a log P of greater than about 1.5.

4. The composition of Claim 1, wherein said aminopolysaccharide is selected from the group consisting of chitosan, chitosan derivatives, amino substituted cellulose, and combinations thereof.

5. The composition of Claim 1, wherein said negatively charged polysaccharide is selected from the group consisting of xanthan, guar gum, carrageenan, pectin, alginic acid, and combinations thereof.

6. The composition of Claim 5, wherein said polysaccharide is xanthan.

7. The composition of Claim 1, wherein said particle comprises greater than about 50% chitosan.

8. The composition of Claim 1, wherein said particle comprises greater than about 60% chitosan.

9. The composition of Claim 1, wherein said particle comprises greater than about 70% chitosan.

10. The composition of Claims 1, wherein said chitosan is a chitosan salt.

11. The composition of Claim 1, further comprising at least one food additive selected from the group consisting of flavorings, colors, vitamins, minerals, fragrances, and phytonutrients.

12. The composition of Claim 1, wherein said composition remains dispersed in a liquid after stirring.

13. The composition of Claim 1, further comprising an anionic compound selected from the group consisting of ribonucleic acid, deoxyribonucleic acid, and anionic drugs.

14. A composition comprising a powder dispersable in a liquid, said powder comprising at least one food additive and particles comprising an aminopolysaccharide and a negatively charged polysaccharide.

15. The composition of Claim 14, wherein said negatively charged polysaccharide is selected from the group consisting of xanthan, guar gum, carrageenan, pectin, and alginic acid.

16. The composition of Claim 14, wherein said polysaccharide is xanthan.

17. The composition of Claim 14, wherein said particle has a partition coefficient of greater than about 1.5.

18. The composition of Claim 14, wherein said particle comprises greater than about 50% aminopolysaccharide.

19. The composition of Claim 14, wherein said particle comprises greater than about 60% aminopolysaccharide.

20. The composition of Claim 14, wherein said particle comprises greater than about 70% aminopolysaccharide.

21. The composition of Claim 14, wherein said aminopolysaccharide is chitosan or a chitosan salt.

22. The composition of Claim 14, wherein said at least one food additive is selected fromb the group consisting of flavorings, colors, vitamins, minerals, fragrances, and phytonutrients.

23. The composition of Claim 14, wherein said composition remains dispersed in a liquid after stirring.

24. A method comprising:

a) providing a positively charged polysaccharide, a negatively charged polysaccharide, and a liquid;

b) combining said positively charged polysaccharide, said negatively charged polysaccharide, and said liquid to form a mixture; and

c) blending said mixture under conditions such that particles containing said positively charged polysaccharide and said negatively charged polysaccharide are formed.

25. The method of Claim 24, wherein said particles bind compound having a partition coefficient of greater than about 1.5.

26. The method of Claim 24, wherein said negatively charged polysaccharide is selected from xanthan, guar gum, carrageenan, pectin, and alginic acid.

27. The method of Claim 24, wherein said negatively charged polysaccharide is xanthan.

28. The method of Claim 24, wherein said particle comprises greater than about 50% chitosan.

29. The method of Claim 24, wherein said particle comprises greater than about 60% chitosan.

30. The method of Claim 24, wherein said particle comprises greater than about 70% chitosan.

31. The method of Claim 23, wherein said chitosan is a chitosan salt.

32. The method of Claim 23, further comprising the step of adding a food additive selected from flavorings, colors, vitamins, minerals, fragrances, and phytonutrients.

33. The particle formed by the method of Claim 23.